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THE REACTION OF CERTAIN CEREALS

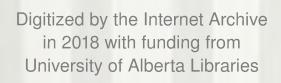
TO FREEZING TEMPERATURES

Arnold William Platt

Department of Field Crops
University of Alberta

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THE REACTION OF CERTAIN CEREALS TO FREEZING TEMPERATURES.

Arnold William Platt
Department of Field Crops.

A THESIS

submitted to the University of Alberta
to fulfil approximately two-thirds of the
requirements for the degree of
MASTER OF SCIENCE

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TABLE OF CONTENTS

	Page
Introduction	1
General methods	4
The effect of freezing temperatures and of defoliation on the subsequent growth of wheat plants	7
Literature review	7
Experiment 1	8
Methods	8
Results	12
Experiment 2	27
Methods	27
Results	28
Differential ability of varieties to recover from defoliation and frost injury in the seedling	
stage	34
Discussion	36
The effect of various factors on the survival of plants exposed to freezing temperatures	39
Soil moisture content	39
Hardening	49
Border effect within flats	54
Endosperm development of the seed	56
The reaction of differently nourished plants to freezing temperatures	59
Literature review	59
Me thods	61
Results	64
Discussion	73

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TABLE OF CONTENTS (continued)

	Page
The reaction of wheat, oat and barley varieties to freezing temperatures	75
Literature review	75
Varietal reactions to freezing temperatures in the seedling stage	76
Methods	76
Results	77
Varietal reactions to freezing temperatures in the heading stage	87
Me thods	87
Results	88
Discussion	92
Summary	95
Acknowledgments	98
References	99

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THE REACTIONS OF CERTAIN CEREALS TO FREEZING TEMPERATURES.

A.W. Platt

INTRODUCTION.

The effect of low temperatures on plant growth has been a subject of interest to man since he first began his study of plant life. As few areas in the world are free from the risk of damage to plant life through the advent of low temperatures, the subject is still of almost world-wide interest. For example, we find reference in the literature to damage by low temperatures suffered by rye in Alberta, by corn in Michigan, by cowpeas in Kansas, by cotton in Mississippi, by sugar cane in Cuba and by coffee in Brazil.

Early investigations upon this subject were greatly hampered as the investigators were obliged to depend upon naturally occurring low temperatures, or on mixtures of ice and salt, or on similar devices, with the attendant difficulties in reaching the desired temperatures and maintaining them for any length of time. In recent years the advent of mechanical refrigeration has removed or greatly lessened these difficulties, and since that time a good deal of progress has been made in the study of plant behavior at low temperatures.

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In cereal crops the work on the low temperature reaction of plant tissue has largely been confined to the so-called fall or winter varieties, probably because these varieties are of necessity exposed to relatively low temperatures and, as a result, the damage sustained is frequently very great. In contrast to this the spring varieties from the standpoint of their reaction to low temperatures, have been largely neglected.

In the present investigation the reactions of spring varieties of wheat, oats and barley to freezing temperatures in the seedling stage and of spring wheat varieties in the heading stage, were studied. The investigation may be said to have had four objectives, namely:

- 1. To determine the effect of exposures to freezing temperatures on the subsequent growth of the plants.
- To determine the relative resistance of plants growing under various nutritional conditions to freezing temperatures.
- 5. To determine the effect of various local environmental factors prevalent during the growth and the freezing, of plants used for experimental purposes, on the frost injury sustained by such plants.
- 4. To determine whether or not varietal differences, with respect to injury by freezing temperatures, exist in the commonly grown varieties of wheat, oats and barley, and to determine as far as possible the magnitude of any such differences.

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The results obtained with regard to each aspect of the investigation referred to above will be presented in separate sections of this paper, together with a brief review of the relevant literature and the methods followed in carrying out the experimental work. THE PERSON NAMED IN COLUMN TWO

GENERAL METHODS

In this investigation a York refrigerating plant was used to obtain the low temperatures desired. Two chambers, each of which was 9 feet by 10 feet by 6.5 feet high, were available for use. The outer chamber was fitted with artificial lights and used as a hardening chamber, while the inner chamber was used for exposing the plants to freezing temperatures. The refrigerating coils in these chambers are placed overhead. Circulation of air to insure uniform temperatures throughout the chamber, was accomplished by means of a Sirocco suction fan and, in addition, by two office fans placed at strategic points within the chamber. Despite these precautions pockets of warm and cold air did occasionally form during the freezing exposure and, in consequence, adequate replication was necessary in order to allow for the variations in damage that resulted from such pocket formation. temperature within the chamber was controlled by means of a thermo-regulator which allowed a variation in temperature of about 1.50c.

Unless otherwise stated, the material was grown in wooden flats the dimensions of which were 22 by 16 by 4 inches.

A black, highly fertile soil, characterized by a high organic matter content and known as Edmonton loam, was used in all experiments unless otherwise noted.

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In many of the experiments the plants were pre-chilled, that is, they were exposed to a temperature of 0°C. just previous to an exposure to freezing temperatures. The duration of the pre-chilling period varied in the different experiments and consequently will be specified in each individual instance.

After an exposure to freezing temperatures the plants were allowed to recover for about ten days before the damage sustained by them was estimated. At that time the number of plants falling into each of four classes was noted. In class one were placed those plants that were killed; in class two those severely injured; in class three those slightly injured; and in class four those that were apparently uninjured. Each plant falling into class one was given a survival value of 0.0, each into class two of 0.33, each into class three of 0.66 and each into class four of 1.0. For a given lot of plants these values were summated, multiplied by 100 and divided by the number of plants present to give a survival index for that particular lot of plants. The survival indices so calculated range from 0 to 100, 0 indicating complete killing and 100 indicating no apparent damage.

Similar survival indices have been found by Martin (8), Salmon (14), Aamodt and Platt (1) and others, to be highly correlated with the percentage of dead plants. The objection to using this latter criterion of injury is the fact that in many experiments no plants whatever are killed, even though there may be marked differences in injury. The percentage of

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plants killed in such cases gives no indication whatever of the degree of injury and consequently when the investigator depends upon this, no results are secured from the experiment. Since it is sometimes difficult to accurately determine, beforehand, the temperature at which differential killing will occur, dependence on percentage kill alone means that a certain portion of the experiments will contribute no data of value.

The analysis of variance as presented by Fisher (2) has been used for the statistical treatment of the data obtained wherever this was possible. However the significance of the mean square of any possible variable, compared with the mean square for error, has been determined by the F method given by Snedecor (17) rather than the Z method given by Fisher.

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The Effect of Freezing Temperatures and of Defoliation on the Subsequent Growth of Wheat Plants

Literature review.

The investigations of Waldron (19,20) appear to be the only work published to date on the effect of frost injury on the subesquent growth of wheat plants.

In his earlier work (19) Waldron reports that the varieties Ceres and Hope were growing in each of two spring wheat nurseries. These nurseries were growing under apparently uniform conditions, except that one nursery received some shelter from a windbreak. A frost occurred which damaged the unsheltered nursery but did not damage the sheltered one. In the former nursery Hope was found to be severely damaged while Ceres exhibited little or no damage. At hervest it was found that the yield of Hope and of Ceres, in the nursery showing no frost damage, was approximately equal but in the nursery showing frost damage, the slightly damaged Ceres outyielded the severely damaged Hope by approximately 13 bushels per acre. Waldron attributes this relative lowering of the yield of Hope to the frost damage sustained by it in the seedling stage:

Later (20) a hybrid nursery was damaged by spring frosts. Waldron noted that in rows of F₅ material, and in the variety Hard Federation, some of the plants were severely injured while others escaped without injury. The injured and non-injured plants were tagged. At maturity it was noted that

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the culms of the injured plants were about 10 per cent shorter than those of the non-injured plants, and that the number of fertile culms and the grain yield per plant was reduced by about 50 per cent. No mention was made as to the relative growth periods of the injured and non-injured plants.

The differences in injury sustained by the individual plants are regarded by Waldron as probably genetical in nature. He points out that should the genes for susceptibility to frost be responsible for, or linked with, the genes for lessened height and lowered yield, the conclusions drawn would not be valid. However, the inclusion of the rows of Hard Federation, which behaved similarly to the hybrid lines, suggested that such an association of genes did not exist.

Waldron concludes that injuries of this sort must have an effect on yield per acre, as any increased growth of the non-injured plants above what would have occurred normally could not compensate for the losses of the injured plants, which would remain as competitors.

Experiment 1.

Me thods.

The first experiment was concerned with a comparison of the growth period, height and number of fertile culms per plant of frozen, defoliated and non-injured plants.

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Five varieties of spring wheat, namely: Garnet C.A.N.*1316, Reward C.A.N. 1509, Red Bobs 222 C.A.N. 1637, Marquis C.A.N. 1621 and Canus, were used.

As the exposure to freezing temperatures had to be made in the freezing chamber, it was necessary to grow the plants in containers in order that they might be moved in and out of the chamber.

The first lot of plants used was grown in six-inch flower pots. Five seeds were planted in each pot and upon emergence the plants were thinned to a uniform stand of three plants per pot.

When the plants reached the -two-leaf stage they were divided into two lots. One lot was again sub-divided into four sub-lots. One sub-lot was left as a non-injured check, two sub-lots were exposed to freezing temperatures, the first at -8°C. for four hours and the second at -12°C. for four hours; and the fourth sub-lot was clipped off about one-half inch above the surface of the soil.

It was desired to injure all varieties to approximately the same degree. In order to attain this objective it was necessary to select plants having the required smount of injury from each of the varieties used. In the lightly frozen lot only plants having less than 50 per cent of their foliage injured were saved, while in the severely frozen lot only plants having

^{*} Canadian accession number.

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more than 50 per cent of their foliage injured were saved.

The appearance of typical lightly and severely frozen plants at the time of selection is shown in Figure 1.

The second lot was treated in the same manner as the first lot when it reached the five-leaf stage, and the frozen plants were again similarly selected.

All this material was allowed to grow to maturity in the greenhouse.

The second group of plants used in the first experiment were grown in flats. These flats were sub-divided into compartments by bottomless cardboard boxes which were two inches square and extended to the bottom of the flat. One plant was grown in each compartment.

The plants were frozen in the two- and five-leaf stages, as described above, and the frozen plants were samilarly selected.

At the time of selection all plants were transplanted to the field into rows one foot apart, and with the plants two inches apart within the row. Because of the cardboard containers it was possible to lift out the plants with the soil surrounding them, and place them in the field without disturbing them to any extent. The only sign of injury observed was in some of the plants transplanted in the five-leaf stage where, in some cases, the roots had run along the bottom of the flat and consequently became broken off in transplanting. In all, over 4,000 plants were so transplanted and, by actual count, there was 100 per cent survival.

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For the purpose in hand, namely the comparison of injured and non-injured plants, the method appeared by careful observation to be quite suitable. The only suggestion of a differential effect noted was that the check lots of plants appeared to suffer more after transplanting than did the defoliated or severely frozen plants, probably as a result of their greater development of foliage. Weather conditions were ideal at the time of transplanting, and this fact probably accounts to a large extent for the good results obtained.

It should be noted that when the plants were grown to maturity in the greenhouse, all plants were growing under similar environmental conditions at all times, but that this was not so when the plants were transplanted to the field, as those injured in the five-leaf stage remained in the greenhouse for some time after those injured in the two-leaf stage had been removed to the field.

The date of heading of each individual plant was recorded.

At maturity the plants were harvested and the number of fertile culms per plant, as well as the height in centimetres of each plant from the base to the apical spikelet excluding awns, was recorded.

The yield per plant was not recorded as the grain from the material grown in the field was so badly damaged by fall frosts that data on yield per plant would have been valueless.

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Results.

Days from emergence to heading. The data obtained with regard to the days from emergence to heading of injured and non-injured plants grown in the field and in the greenhouse, are presented in Table I. For convenience the differences in days from emergence to heading between each mean, and all other comparable means, were calculated. These data are presented in Table II.

It will be noted that in all cases the injured plants were later in heading than were the non-injured plants and that these increases are statistically significant. Amongst the injured groups those severely frozen were in general the latest in heading. In all cases they were later than the lightly frozen plants and, except for the Marquis and Canus plants injured in the two-leaf stage and grown in the field, they were significantly later than the defoliated plants. On the average the defoliated plants were later than the lightly frozen plants, but in many individual instances this was not the case, particularly amongst plants injured in the two-leaf stage and grown in the greenhouse.

It will be noted that, on the whole, the differences between groups, especially the differences between the non-injured and the injured groups, were accentuated when the plants were grown to maturity in the greenhouse as compared with the differences obtained when the plants were grown to maturity in

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TABLE I

Days from emergence to heading of wheat plants injured in the seedling stage by frost and by defoliation.

Stag t wh	Stage at which plants were	Type of	Green No. of	ys from	m emergenc	Days from emergence to heading enhouse Fi	r i e i d	e E
	inja		plents	Mean	of mean	plents	Mean	of mean
2-leaf stage No injury Light frost Foliage rem Severe fros	No inj Light Foliae Severe	No injury Light frost Foliage removed Severe frost	0 8 8 8 4 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	36.17 40.69 41.37 47.82	0.84 0.44 0.65	102 55 88	44.34 45.48 51.05 53.59	0000
5-leaf stage No injury Light frost Follage remo Severe frost	No inj Light Foliag Severe	No injury Light frost Follage removed Severe frost	8855 875 875 875 875 875 875 875 875 875	36.17 41.96 45.40 50.00	0.26	8 8 8 9 0 9 0 0	40.40 41.26 44.36 51.85	0.13 0.32 0.52 0.80
2-leaf stage No injury Light frost Foliage remo Severe frost	No injuitelight i	No injury Light frost Foliage removed Severe frost	59 30 49	35.44 41.21 40.63 47.12	0.16 0.39 0.40 0.56	26 107 90	44.11 45.99 49.39 51.06	0000 8.000 4.000
5-leaf stage No injury Light frost Foliage remo Severe frost	No injuit 1 Light 1 Foliage Severe	No injury Light frost Foliage removed Severe frost	59 30 35 35 35	35.44 42.13 42.67 47.11	0.16 0.31 0.59	78 8 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4	41.21 42.54 46.96 49.51	0.13 0.31 0.55
2-leaf stage No injury Light frost Foliage remo Severe frost	No inju Light i Follege Severe	No injury Light frost Foliage removed Severe frost	60 36 30 47	57.92 42.19 42.77 49.79	0.19	125 64 40 40 40 40	49.89 51.56 52.05 55.63	0.000

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TABLE I (continued)

	Stake			Days f	Days from emergence to heading	ence to be	sading	
	at which		GFB	e n	u s e		F 1 e 1	
Variety	plants were injured	Type of injury	No. of plents	Meen	S.E.	No. of plants	Mean	S.E. of mean
Red Bobs 222 (continued)	5-leaf stage	No injury Light frost Follage removed Severe frost	80 80 84 84	37.92 41.82 43.77 51.92	0.19 0.23 0.98	58 28 14	46.84 48.25 52.00 54.86	0.34 0.25 0.75
Merquis	2-leaf stage	No injury Light frost Foliage removed Severe frost	9 th th th	43.00 44.97 48.07 52.62	0.80 0.67 1.09	31 64 64	52.48 54.59 57.67	0000
	5-leaf stage	No injury Light frost Foliage removed Severe frost	0 4 E	45.00 46.82 48.77 56.17	0.20 0.47 0.42 1.03	67 79 41	51.73 53.42 57.63 58.98	0.19 0.28 0.45
Cenus	2-leaf stage	No injury Light frost Foliage removed Severe frost	8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8	43.12 46.08 47.33 51.10	0.12 0.63 0.55	2 2 2 4 4 4 5 5 5 5 5 5 5 5 5 5 5 5 5 5	50.56 55.54 56.09	0.31 0.15 0.37 0.28
	5-leaf stage	No injury Light frost Folisse removed Severe frost	62 28 4 4	43.12 46.23 49.89 53.14	0.12 0.33 0.38 1.97	88 88 88 88 88 88 88 88 88 88 88 88 88	49.18 52.51 54.61 58.13	0.33 0.26 0.44 0.77

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The difference in days from emergence to heading between the mean of any group of plants and all other comparable means.

•		Stage		Differe (Hor	Direction in days from emergence to heading (Horizontal column.)	lumn-Vert	ergence ical colu	to heading	1
2-leaf stage 2-leaf stage 5-leaf stage 5-leaf stage 5-leaf stage 5-leaf stage		at which		9 H	Greenhouse	6 0	ŝe,	e 1 d	
2-leaf stage 5-leaf stage 5-leaf stage 5-leaf stage 5-leaf stage 5-leaf stage	Variety	plants were injured	Type of injury	Light	Foliage	Severe	Light	Foliage S removed	Severe
2-leaf stage 5-leaf stage 5-leaf stage 5-leaf stage	Garnet	2-leaf stage	No injury Light frost Foliage removed	+4.52*	+5.20	+11.65 + 7.13 + 6.45	+1.14	+6.71	+9.25 +9.11 +2.54
2-leaf stage 5-leaf stage 5-leaf stage		5-leaf stage	No injury Light frost Foliage removed	+2.79	+9.23	+13.83 + 8.04 + 4.60	+0-86	+3.96 +3.10	+11.45 +10.59 +7.49
5-leaf stage 2-leaf stage 5-leaf stage	Reward	2-leaf stage	No injury Light frost Foliage removed	+5.77	+5.19	+11.68 + 5.91 + 6.49	+1.88	+5.28	+ 6.95 +5.07 +1.67
2-leaf stage 5-leaf stage		5-leaf stage	No injury Light frost Foliage removed	+6.69	+7.23	+11.67 + 4.98 + 4.44	+1.33	+5.75	+8.30 +6.97 +2.55
	Red Bobs 222	2-leaf stage	No injury Light frost Foliage removed	+4.27	+4.85	+11.87 + 7.60 + 7.02	+1.67	+2.16	+5.74 +4.07 +3.58
		5-leaf stage	No injury Light frost Foliage removed	+3.90	+5.85	+14.00	+1.41	+5.16	2.86

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5/1	Stage		Difference	liference in days from emergence to beading	from emerg	ence to	Beading	
Variety	at which plants were injured	Type of injury	Gree Light frost	Greenhouse Light Foliage Se frost removed f	Severe frost	Light frost	F 1 e 1 d Light Follage Severe frost removed frost	Severe
Marquis	2-leaf stage	2-leaf stage No injury Light frost Foliage removed	+1.97	+5.07 +3.10	+ + 4.55	+2.11	+5.19	+5.54
	5-leaf stage	No injury Light frost Foliage removed	+3.82 +3.82	+5.77	+13.17 + 9.35 + 7.40	+1.69	+5.90	+7.25 +5.56 +1.35
Canus	2-leaf stage	No injury Light frost Foliage removed	+2.96	+4.21	+ + 5.02 + 3.77	+2.78	+4.97	+5.53 +2.75 +0.56
	5-leaf stage	No injury Light frost Foliage removed	+3.11	+6.77	+10.02	+3.33	+5.43	+8.95 +5.62 +3.52

*Underlined values indicate that these differences exceed twice the standard error of the difference.

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the field. However, the relative differences between groups grown in the greenhouse and comparable groups grown in the field is quite consistent.

In comparing the degree of retardation in date of heading sustained by plants injured in the two, and those injured in the five-leaf stage, it will be noted that for the most part those injured in the five-leaf stage were, relative to the checks, about one to two days later than those injured in the two-leaf stage.

Height. The data obtained on the height of injured and non-injured plants grown in the field and in the greenhouse are presented in Table III. For convenience, the differences in height between each mean and all other comparable means were calculated. These data are presented in Table IV. From the data presented it will be seen that, in the majority of instances, the differences noted between the mean heights of any two comparable groups of plants are less than 10 centimetres in extent, and that only two of these exceed 14 centimetres.

It will also be noted from the data presented that no apparent relationship, for all varieties and conditions present, exists between height at maturity and injury in the seedling stage. There is a tendency for the frozen and defoliated plants to be shorter than the non-injured plants when they were grown to maturity in the greenhouse, with the exception, particularly, of Garnet plants. The lightly frozen plants when grown in the

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TABLE III

Height in centimetres at maturity of wheat plants injured in the seedling stage by frost and by defoliation.

	40.00			Ħ	Height in cms.	cms.		
	at which		0	enho	use		F 1 6 1	P
Variety	plents were injured	Type of injury	l	Mean of mean	S.E. of mean	No. of plants	Mean	S.E.
Garnet	2-leaf stage	No injury Light frost	57	67.63	1.65	32	82.77	
		Foliage removed Severe frost	26	68.44	2.03 40.03	8 22	82.14	1.45
	5-leaf stage	No injury Light frost	18	67.65	1.00	58	72.06	
		Foliage removed Severe frost	128	75.01 70.00	3.14	36	59.82	1.70
		1000		90	00	96	, 40	
Keward	asers Teat-7	Light frost	0 G	71.79	1.15	108	92.55	0.72
		Foliage removed	63	61.77	1.42	46	78.45	
		Severe frost	49	61.29	1.50	22 00	83.49	
	5-leaf stage	No injury	09	75.00	1.08	88	69.81	
)	Light frost	30	60.99	1.49	98	72.42	0.78
		Foliage removed	83	55.86	1.54	45	66.12	
		Severe frost	30	65.61	1.86	5	66.84	
Red Bobs	2-10af stape	No in inra	09	81.34	1.32	28	92.46	
222		Light frost	63 CD	78.82	1.69	125	90.45	1.01
		Foliage removed	30	71.59	1.59	64	99.24	
		Severe frost	37	76.57	1.56	40	92.19	1.51
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						of Toles	<u>2</u>

TABLE III (continued)

	Stage at which		6 8-		Height in cms.			
Variety	plants were injured	Type of injury	No. of plants	Mean of mea	S.E.	No. of plants	Mean	S.E.
222 (continued)	5-leaf stage	No injury Light frost Foliage removed Severe frost	60 30 17 17	81.34 75.13 73.09	11.22 20.74 20.04 00.00	59 30 14	81.42 81.12 79.50	1.12
Marquis	2-leaf stage	No injury Light frost Foliage removed Severe frost	0 4 8 0 0 4 0 0	86.41 82.20 82.99	1.082	31 148 66	95.52 99.64 94.05	1.55
	5-leaf stage	No injury Light frost Foliage removed Severe frost	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	86.41 86.89 82.40	1.02 2.1.08 2.1.1.1.1.1.1.1.1.1.1.1.1.1.1.1.1.1.1.1	0 6 4 4 0 6 8 8	72.27 79.26 76.35 82.26	1.01
Canus	2-leaf stage	No injury Light frost Foliage removed Severe frost	60 83 88 88	84.56 82.70 86.20 75.00	1.10 1.44 1.48	134 46 65	87.87 98.22 90.12	2.01 0.62 1.28 0.94
	5-leaf stage	No injury Light frost Foliage removed Severe frost	12400	84.56 82.64 80.90 76.07	1.11 0.79 1.76 2.67	100 37 32	81.06 76.77 80.43 78.39	1.02

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	1919	, L , 9 %			45.86		44 10 10 10 10 10	
7853		eage.	* * * *		机 意用 图			

TABLE IV

The difference in height at maturity between the mean of any group of plants and all other comparable means.

column) Severe frost	- 3.72 - 13.59 - 3.09	- 3.06 + 9.18 + 9.18	2.67 9.06 5.04	5.58	1.74	3.33
Difference in height (Horizontal column-vertical column Greenhouse Field Light Foliage Severe Light Foliage Severe frost removed frost	0.63	12.24	-14.10	2.69	+ 6.78	1.62
tal column Fight frost	+9.87	+1.68	+6.39	+2.61	-2.01	-0.30
(Horizon u s e Severe frost	+8.61 -1.35 +7.80	+2.37 -7.65 -5.01	-13.71 -10.50 - 0.48	9.39	2.25	- 4.11 + 2.10 + 4.14
ence in height (Hori Greenhouse Ght Follage Seven	+0.81	+7.38 -2.64	13.23	19.14	9.75	8.25
ference Gre Light frost	+ 9.96*	+10.02	- 3.21	- 8.91	83 83 -	- 6.21
Type of injury	No injury Light frost Foliage removed					
Stage et which plants were injured	2-leaf stage	5-leaf stage	2-leaf stage	5-leaf stage	2-leaf stage	5-leaf stage
Variety	Garnet		Reward		Red Bobs 222	

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(continued)

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The difference in height at maturity between the means.

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	Stage		Difference in height (Horizontal column-vertical column)	in height	(Horizo	ntal colu	mn-vertice	1 column)
	at which		Gr	Greenhouse	use	FI	F 1 6 1 d	
Variety	plants were	Type of	Light	Foliage Severe	Severe	Light	Foliage	Severe
	in jured	injury	frost	removed	frost	frost	removed	frost
Merquis	2-leaf stage	No injury Light frost Foliege removed	-3.21	- 3.42	-4.05 -0.84 -0.63	+4.12	-0.47	-2.16 -6.28 -0.69
	5-leaf stage	No injury Light frost Foliage removed	+0.48	4.49	-9.09 -9.57 -5.08	+6.99	-2.91 -2.91	- 21
Cenus	2-leaf stage	No injury Light frost Foliage removed	-1.86	+1.64	-9.56 -7.70 -11.20	+10.35	-8.10	+9.57
	5-leaf stage	No injury Light frost Foliage removed	-1.92	-3.66	6.57 -6.57	-4.29	-0.63	-2.67 +1.62 -2.04

*Underlined values indicate that these differences exceed twice the standard error of the difference.

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field show a tendency to be taller than the non-injured plants. The defoliated and severely frozen plants grown in the field failed to show any tendency to be generally shorter or taller than the comparable non-injured plants.

Fertile culms per plant. The data obtained on the number of fertile culms per plant of plants grown in the field and in the greenhouse are presented in Table V. For convenience the differences in the number of fertile culms per plant between each mean and all other comparable means were calculated. These data are presented in Table VI.

From the data presented it will be noted that when the plants were grown to maturity in the greenhouse there was, for the most part, little difference in the mean number of fertile culms per plant between any of the groups studied. As three plants were grown in each six-inch flower pot, it is believed that one or more soil nutrients became limiting to such an extent that this factor was of greater importance in determining the number of culms per plant than were the treatments to which the plants were subjected. This belief is strengthened by the fact that varietal differences between the means of check lots are practically non-existent under these conditions, while under normal conditions such differences would be expected and were, in fact, observed when the plants were grown to maturity in the field.

According to the data obtained, the defoliated and severely frozen plants grown in the field had, with the exception of the defoliated Reward plants injured in the two-leaf stage, and

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TABLE V

Number of fertile culms per plant of wheat plants injured in the seedling stage by frost and by defoliation.

	e9.678			Fert	Fertile culms per plant	per plant			
	at which		G r e	e n h o	u s e		Fiel	p	
Variety	plants were injured	Type of injury	No. of plants	Mean	S.E. lean of mean	No. of plants	Mean	S.E.	
Garnet	2-leaf stage	No injury Light frost Foliage removed Severe frost	300 31 31	8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8	0.12 0.19 0.16 0.18	32 102 55 88	3.00 3.13 15 15 15 15 15 15 15 15 15 15 15 15 15	0.22 0.19 0.19 1.0	- 2
	5-leaf stage	No injury Light frost Foliage removed Severe frost	59 26 26 17	2.02 2.57 2.92 2.92	0.00 0.00 44 48	58 71 36 40	8888 8888 8889	0.17	3 -
Reward	2-leaf stage	No injury Light frost Foliage removed Severe frost	0 8 8 8 4	22.98 22.44 1.834 1.80	0.11 0.20 0.21 0.14	26 107 46 85	44.8 118.6 118.8 118.8 118.8	0.32 0.16 0.12 0.12	
	5-leaf stage	No injury Light frost Foliage removed Severe frost	38890	2.59 2.59 1.84	0.11 0.16 0.22 0.19	88 88 84 84 85 85 88	8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8	0.13	
Red Bobs 222	2-leaf stage	No injury Light frost Foliage removed Severe frost	88884 8408	1.50	0.08 0.12 0.17 0.13	28 125 64 40	6.18 5.17 3.80	0.33 0.16 0.21 0.23	

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TABLE V (continued)

	Stage			Fert	Fertile culms per plant	per plant		
	at which		0	e n h	enhouse		F 1 e 1	đ
Variety	plants were injured	Type of injury	No. of plants	Mean	S.E.	No. of plants	Mean	S.E.
Red Bobs 222 (continued)	5-leaf stage	No injury Light frost Foliage removed Severe frost	8 2 2 2 2	1.64 1.64 1.36	0.08 0.09 0.13	59 117 30 14	4 4 5 5 5 4 5 6 4 5 6 4 5 6 4 5 6 4 5 6 4 5 6 4 5 6 4 5 6 4 5 6 4 6 6 4 6 6 4 6 6 6 4 6 6 6 6	0.23 0.28 0.26
Marquis	2-leaf stage	No injury Light frost Foliage removed Severe frost	50 kg 950 50 50 50 50 50 50 50 50 50 50 50 50 5	8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8	0.15 0.15 0.15	31 138 66 60	40.55 44.55 44.55	0.40 0.20 0.17
	5-leaf stage	No injury Light frost Foliage removed Severe frost	1 2 2 1 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2	2.04 2.04 1.42	0.13 0.13 0.24	67 79 41 43	4.19 5.19 5.85 5.55	0000
Cenus	2-leaf stage	No injury Light frost Foliage removed Severe frost	3350	2.27	0.09 0.13 0.13	134 466 655	7.74 6.06 4.96 5.11	0000
	5-leaf stage	No injury Light frost Foliage removed Severe frost	000 000 14	2.17 1.98 2.34 1.86	0.09 0.09 0.18 0.23	100 37 32	4.92 4.17 4.11 3.78	00.00 0.00 0.00 0.00 14.00

The differences in fertile culms per plant between the mean of any group of plants and all other comparable means.

	S. C. B. C.	А	ifferenc	e in no.o	f fertile	culms	Horizontel	Difference in no. of fertile culms (Horizontel column-vertical column)
	at which		H	eenhouse	n s e	Se.	1610	П
Variety	plants were injured	Type of injury	frost	removed	frost	frost	removed	frost
Garnet	2-leaf stage	No injury Light frost Foliage removed	60.0-	+0.81*	+0.53	+1.74	-0.96	-0.54 -0.42 +0.42
	5-leaf stage	No injury Light frost Follage removed	+0.55	+0.90 +0.35	+0.10 -0.45 -0.80	-0.24	-1.22	1.25
Reward	2-leaf stage	No injury Light frost Foliage removed	-0.57	-0.64	-1.18 -0.61 -0.54	+0.66	-0.52	-1.30 -1.96 -0.78
	5-leaf stage	No injury Light frost Follage removed	-0.39	-0.08	-1.14 -0.75 -1.06	-0.04	-0.79	-0.91 -0.87 -0.12
Red Bobs	2-leaf stage	No injury Light frost Foliage removed	-0.05	-0.14	+0.08	-1.25	-1.01 +0.24	-2.38 -1.13
	5-leaf stage	No injury Light frost Foliage removed	0.0	+0.03	-0.28 -0.28 -0.31	-0.14	-0.91	-1.78 -1.64 -0.87

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TABLE VI (continued)

	Stage		Ä H	Difference in no. of fertile culms (Horizontal column)	in no. o	f ferti	column)	
	et which		Gr e	Greenhouse	1 8 6	E	1 0 1 0	
Variety	plants were injured	Type of injury	Light	Foliage removed	Severe	Light	Light Foliage frost removed	Severe
Marquis	2-leaf stage	No injury Light frost Foliage removed	-0.20	-0.07 +0.13	+0.06 +0.26 +0.13	+0.67	-1.23	-1.64 -2.31 -0.41
	5-leaf stage	No injury Light frost Foliage removed	-0.23	-0.20	0.85	66.00	-0.34	+0.37 -0.62 +0.71
Cenus	2-leaf stage	No injury Light frost Foliage removed	+0.10	-0.20	-0.14 -0.24 +0.06	-1.68	-2.78	-2.63 -0.95 +0.15
	5-leaf stage	No injury Light frost Foliage removed	-0.19	+0.17	-0.10 -0.48	-0.75	-0.81	-1.14 -0.39 -0.33

*Underlined values indicate that the differences noted exceed twice the standard error of the difference.

TABLE VI (continued)

30	2.75 S.78	TO.0+ 48.0- 02	1.00 C.1.	Licer Cones Licer
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	<u>(1)</u>		C. F.	Ctoluny

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the defoliated and severely frozen Marquis plants injured in the five-leaf stage, a smaller number of fertile culms per plant than did the comparable non-injured plants.

Considering the lightly frozen plants, it will be noted that those of Garnet injured in the two-leaf stage had more fertile culms per plant than did comparable non-injured plants; while those of Red Bobs 222 injured in the two-leaf stage and those of Canus injured in the two- and the five-leaf stage had, conversely, fewer fertile culms per plant than the comparable non-injured plants; and that in all other cases the differences noted are not statistically significant.

The diminution in number of fertile culms per plent sustained by the injured plants in comparison with non-injured plants did not appear to be consistently influenced by the stage of development at which the injury occurred.

Experiment 2.

Methods.

The second experiment was concerned with a comparison of defoliated and normal plants grown under field conditions.

The five varieties, used in the first experiment, were grown in five-row plots, the rows being 18 feet long and one foot apart.

The young plants were subjected to three treatments.

One lot was left as the non-injured check, a second was defoliated at the two-leaf stage, and a third was defoliated at the five-leaf

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stage. Each treatment was replicated four times.

The height at maturity, growth period and yield of each plot were recorded.

Results.

Growth period. A summary of the data obtained on the growth periods of defoliated and non-injured plants grown under field conditions is presented in Table VII. An analysis of variance was calculated from the data obtained, and is presented in Table VIII.

According to the data presented the defoliated plants were later in maturing than were the non-injured plants. the average those plants defoliated in the two-leaf stage were about three days, and those defoliated in the five-leaf stage about five days later in maturing than were the non-injured plants. The standard error of the difference between the means of any two treatments was calculated and found to be equal to 0.71 days, consequently differences in excess of 1.4 days may be considered significant. It is evident that the increases in the growth periods of defoliated plants are statistically significant. Also, it is evident that the growth periods of plants defoliated in the five-leaf stage are significantly greater than are those of plants defoliated in the two-leaf stage. This retardation in the maturing of defoliated plants grown under field conditions is but slightly less than the retardation in heading of the defoliated plants grown in the greenhouse or of those transplanted to the field.

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TABLE VII

Height, growth period and yield of normal and defoliated wheat varieties.

	Ħ	Height in cms.		Grow	Growth period in days	n days	Yield	Yield in bus. per acre	r acre
Variety	Normal	Defoliated Defoliated in the in the 2-leaf 5-leaf stage stage	Defoliated in the 5-leaf stage	Normal	Defoliated Defoliated in the in the 2-leaf 5-leaf stage stage	Defoliated in the 5-leaf stage	Normal	Defoliated in the 2-leaf stage	Defoliated Defoliated in the 2-leaf 5-leaf stage
Garnet	109.8	111.3	100.5	95.0	98.3	100.0	40.0	80.00 50.00	40.3
Reward	110.3	111.3	105.8	0.46	100.5	101.0	36.8	33.3	30.9
Red Bobs	116.5	108.0	106.8	0.66	101.3	102.5	44.0	38.3	36.0
Marquis	119.3	110.8	111.3	102.0	103.8	108.0	38.9	36.3	37.4
Canus	115.5	103.8	107.5	102.3	105.0	108.8	42.5	37.9	35.3
Average 114.	114.5	109.0	106.4	99.1	101.8	104.1	40.4	37.0	36.0

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TABLE VIII

Analysis of variance of the growth periods of defoliated and normal wheat varieties.

Variation due to:	egrees of freedom	Sum of squares	Mean square	F
Varieties	4	525.78	131.45	26.24*
Treatments	2	240.60	120.30	24.01*
Varieties x treatments	8	25.02	3.13	-
Blocks	3	5.40	1.80	-
Error	42	210.40	5.01	-
Total	59	1007.20	-	-

^{*} Exceeds the 1% point.

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TABLE IX

Analysis of variance of the height at maturity of normal and defoliated wheat varieties.

Variation due to:	Degrees of freedom	Sum of squares	Mean square	F	
Varieties	4	290.33	72.59	3.29**	
Treatments	2	646.70	323.35	14.65*	
Varieties x treatments	8	388.47	48.56	2.20**	
Blocks	3	190.60	63.53	2.88**	
Error	42	926.90	22.07	-	
Total	59	2443.00	-	-	

^{*} Exceeds the 1% point.

^{**} Exceeds the 5% point.

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TABLE X

Analysis of variance of the yield of normal and defoliated wheat varieties.

	Degrees of	Sum of	Mean	
Variation due to:	freedom	squares	square	F
Varieties	4	280.60	70.15	14.58*
Treatments	2	218.27	109.14	22.69*
Varieties x treatments	8	119.98	15.00	3.12*
Blocks	3	77.63	25.88	5.38*
Error	42	202.02	4.81	-
Total	59	1104.10	_	_

^{*} Exceeds the 1% point.

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***	9	C.F. 8 N.C.L	1;	1-56

Height. A summary of the data obtained on the height of defoliated and non-injured plants is presented in Table VII. An analysis of variance was calculated from the data obtained and is presented in Table IX. The mean square due to treatments is significant and, therefore, it may be concluded that defoliation resulted in a reduction in height.

Yield. A summary of the data obtained on the yield of defoliated and non-injured plants is presented in Table VII.

An analysis of variance was calculated from the data obtained, and this is presented in Table X.

According to the data in Table VII, on the average, those plants defoliated in the two-leaf stage suffered a decrease in yield of 3.4 bushels per acre, and those defoliated in the five-leaf stage suffered a decrease of 4.4 bushels per acre when compared with non-injured plants. The standard error of the difference between the means of any two treatments was calculated and found to be equal to 0.693 bushels, consequently differences in excess of 1.4 bushels may be considered significant. It is evident that the decreases in yield, due to defoliation, are statistically significant. Also it is evident that the yield of plants defoliated in the five-leaf stage was not significantly different from that of plants defoliated in the two-leaf stage.

Unfortunately data on the yield per plant of those plants transplanted to the field are not available for comparison with these results. It will be recalled, however, that in the

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 transplantation experiment defoliation, in almost every instance, resulted in a reduction in the number of fertile culms per plant. If it is assumed that yield is correlated with the number of fertile culms, then the results obtained in these two experiments are essentially similar.

Differential ability of varieties to recover from defoliation and frost injury in the seedling stage.

In these experiments the varieties used suffered approximately the same degree of frost injury in the seedling stage. It was found that such injury modified the maturity, height and number of fertile culms per plant. The question as to whether or not varietal differences occurred in the degree of modification will now be considered.

The data obtained from the comparison of defoliated and non-injured plants in Experiment 2 are of interest in this regard. The significance of the mean squares obtained for the interaction of varieties and treatments (Tables VIII, IX and X) may be used as a measure of differential varietal response. In the cases of yield and height significant interactions were evident (Tables IX and X). It is apparent from the data presented in Table VII that a differential response in yield occurred when the varieties were defoliated at the five-leaf stage, as in this group Garnet and Marquis gave relatively

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higher yields than did Red Bobs 222, Reward or Camus. In other words, the results show that under the conditions of this experiment Carnet and Marquis were better able to recover after defoliation in the five-leaf stage than were the other varieties in the test. Similarly, the data concerning height show that the relative damage resulting from defoliation in the two- and in the five-leaf stages was more severely damaged when defoliated in the five-leaf stage, while Canus suffered more in the two-leaf stage.

Considering the data obtained in Experiment 1, it will be noted that in many individual instances a given variety was apparently better able to recover from injury than were the other varieties. When these data are considered as a whole, however, it is apparent that no one of the varieties was consistently able to recover from the same degree of injury to a markedly greater extent than were any of the other varieties.

From these results it appears that, in as far as seedling frost damage affects the resulting plant growth, the variety showing the greatest seedling injury will have its subsequent growth affected to the greatest extent. It follows, therefore, that seedling frost reaction of wheat plants is a good index of such plants. These conditions are, however, subject to the limitations of these experiments and might need altering if data were available for a greater number of varieties studied for other growth characters under different environmental conditions.

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Discussion.

As has been pointed out, Waldron (20) concluded from the results he obtained that seedling frost injury must reduce the yield per acre. The results obtained in the present investigation are not conclusive in this regard but seem, for the most part, to support this contention. In practically all cases the plants severely injured by low temperatures had fewer fertile culms per plant than did the comparable non-injured plants. Waldron's data show that the yield per culm of the injured plants was lower than the yield per culm of the non-injured plants. If such is the case in the present investigation, the decrease in yield per plant would be greater than the decrease in the number of fertile culms per plant would indicate. The reduction in yield per plant under the conditions of this experiment does not necessarily mean that a reduction in yield per acre would occur with a similar amount of injury under field conditions. when the amount of nutrients per plant was extremely limited, as when the plants were grown to maturity in pots, the majority of the injured plants failed to show a reduction in the number of fertile culms, when compared with the non-injured plants. condition might be expected to occur under certain field conditions. On the other hand a definite reduction in yield per acre was obtained under field conditions due to defoliation. Furthermore, this reduction was at least somewhat comparable to the reduction in fertile culms per plant of the defoliated plants growing under similar conditions to those severely frozen. These latter results

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appear to support the contention of Waldron that frost injury in the seedling stage does cause a reduction in yield per acre under field conditions.

The results obtained with regard to the influence of seedling injury on the resulting plant height indicate that any differences in height, attributable to this factor, are not of sufficient magnitude to be of any significance in commercial wheat production. The great variability obtained in the differences in height between injured and non-injured plants is evidence that, in addition to seedling injury, other factors were exerting a marked influence on this character.

spears to definitely increase the growth period of the plants so injured. In areas such as west central and northern Alberta this lengthening of the growth period is of paramount importance in that it increases the hazard of fall frosts which severely reduce both the yield and the quality of the grain. The data obtained indicate that an increase of about nine days in the growth period may be expected when plants are severely frozen. Such an increase would, in many years, in such areas as have been mentioned, doom the crop to exposure to fall frosts previous to maturity with all the loss that this occasions, aside from any loss in yield that the seedling injury in itself may have caused. It is recognized that the delay observed in the maturing of injured plants in these experiments may not be comparable with that obtained in the field. The only check

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available is that between the defoliated plants growing under field conditions and those growing under conditions similar to the frosted plants. In this comparison the delay occasioned in each instance was of approximately the same duration.

It should be kept in mind that in all cases the injured plants in these experiments developed under conditions very favorable to recovery. Had less favorable conditions prevailed, the results might have been vastly different.

Nevertheless it seems probable that had conditions been less favorable, due to drought or similar factors, the injured plants would have suffered as much or possibly more, relative to the checks, than they actually did.

In conclusion, it may be said that the results obtained support the thesis that damage to wheat seedlings by exposures to freezing temperatures results in injuries that persist throughout the lives of the plants.

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The Effect of Various Factors on the Survival of Plants
Exposed to Freezing Temperatures

Salmon (14) says that success in evaluating the results of artificial freezing and in studying resistance to cold in general, depends on good technique, that is in avoiding various sources of error which may interfere with the logical interpretation of the results obtained. At the beginning of this investigation several experiments were conducted in order to find, if possible, and evaluate what might be sources of error in conducting future experiments using artificially produced low temperatures.

Soil moisture content.

Literature review.

A great many observations have been recorded on the relative cold resistance of plants growing in dry and wet soils. These observations, while of considerable significance from many standpoints, are not of much value in predicting what will happen in artificial tests because there are associated factors, such as heaving, at work in the field that are not operative in these latter tests.

Klages (6) has, however, conducted experiments to determine the relative survival of plants, growing in wet and

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dry soils, when exposed to artificially produced freezing temperatures. He found that plants growing in dry soil were more resistant than those growing in wet soil during the early part of the exposure; but, when killing set in, it progressed much more rapidly among the former than among the latter plants. He believes that the early resistance, shown by the plants grown in dry soil, was due to the retardation of plant growth; while the later resistance shown by the plants grown in wet soil, was due to the protective effect of water. The specific heat of water is 1.000; of sand, 0.193; of clay, 0.206 and of loam, 0.215; consequently the more water a soil contains the slower it will cool. Apparently no attempt was made to separate the effects of the two factors believed to be at work, namely, the physiological effect of varying moisture on the growth of the plant and the physical effect of the protective action of water as such.

Salmon (14), in testing the reaction of winter wheat varieties to artificially produced freezing temperatures, noted that they were more resistant when grown in wet soil. However, when the plants were subjected to prolonged chilling previous to an exposure to freezing temperatures, these differences disappeared. He therefore concludes that the differences originally noted were due to a temperature lag in the wet soil.

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Experiment 1.

Methods.

In this experiment the plants were grown in soil maintained at a uniform moisture content during the growth of the plants, but at varying moisture contents during the exposure to freezing temperatures. Six varieties of spring wheat were used. These were grown in wooden flats, all varieties occurring once in each flat.

All flats were kept at a uniform moisture content (approximately 50 per cent of the moisture-holding capacity of the soil) until just before the plants were subjected to freezing temperatures. At this time they were allowed to dry until the moisture content was 20 per cent of the moisture-holding capacity of the soil. Twelve of the 36 flats were left at this moisture content, 12 were brought to a moisture content of 50, and 12 to a moisture content of 65 per cent of the moisture holding capacity of the soil.

The 36 flats were treated in the following manner. Three lots, each consisting of four flats at 20, four at 50 and four at 65 per cent moisture, were subjected to freezing temperatures. The first was subjected to a temperature of -12°C. for four hours without any pre-chilling; the second was pre-chilled for four hours and the temperature was then lowered to -12°C. and maintained for three hours; the third was pre-chilled for twelve hours and then subjected to a temperature of -12°C. for four hours.

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Results.

A summary of the data obtained in this experiment with regard to the survival of plants growing in soil of varying moisture contents at the time of freezing is presented in Table XI. An analysis of variance was calculated from the data obtained and is presented in Table XII.

It will be noted that, on the average, there is a direct relationship between the survival indices and the moisture contents of the soil. The highest survival indices were obtained when the moisture content of the soil was highest, the lowest when the moisture content was lowest, and intermediate when the moisture content was intermediate.

It will be noted (Table XII) that, neither the mean square due to the first order interaction of varieties and soil moisture contents, nor the mean square due to the second order interaction of varieties and soil moisture contents and freezing treatments, can be regarded as being significant. It can be concluded, therefore, that varying the moisture content of the soil and varying the amount of pre-chilling, did not significantly alter the relative varietal reaction to freezing temperatures.

The mean square due to the interaction of moisture contents and freezing treatments (Table XII) may be regarded as significant. This indicates that the reduction in the survival of plants, due to lower moisture contents, was

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TABLE XI

Survival indices of spring wheat varieties when exposed to freezing temperatures in soil of various moisture contents, and with various periods of pre-chilling.

				7	Mean		FVI	Val	survival index	HON			
		65%	moist	capacity	65% moisture holding capacity	20%	capa	50% moisture holding capacity	ding	20%	20% moisture holding capacity	isture hol	guip
Variety	C.A.N.	V.	Д	ပ	YA.	Ā	Д	ပ	AV.	A	В	υ	AV.
Red Bobs 222	1637	62.5	59.0	33.5	62.5 59.0 33.5 51.7		59.5	29.0	18.8 59.5 29.0 35.8	48.5	48.5 39.6	89.8	39.3
Canus		29.8	0.99	42.8	49.5	30.0	68.3	34.3	44.2	15.0	41.3	21.5	25.9
Garnet	1316	27.5	62.0	25.0	62.0 25.0 38.2	19.0	54.8	19.0 54.8 23.0	32.3	41.8	27.8	19.3	29.6
Marquis	1621	46.3	39.5 30.3	30.3	38.7	24.0	51.3	51.5 52.8	36.0	10.0	33.5	23.3	22.3
Caesium 0.111	1256	30.5	65.3	14.0	36.6	24.5	59.0	59.0 11.5	31.7	17.3	49.5	12.3	26.4
Reward	1509	26.8	41.8 32.0	32.0	33.5	25.0	43.8	25.0 45.8 28.8	32.5	18.3	33.3	21.0	24.2
Average		38.9	55.6	29.6	55.6 29.6 41.4	25.6	56.1	26.6	25.6 56.1 26.6 35.4	25.2	37.5	37.5 21.2	28.0

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Pre-chilled for twelve hours, subjected to a temperature of -12°C. for four hours. 5

⁻ Not pre-chilled. Subjected to a temperature of -12°C. for four hours.
- Pre-chilled for four hours, temperature lowered to - 12°C. and maintained at this for three A - Not pre-chilled.

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TABLE XII

Analysis of variance of the survival indices of wheat seedlings exposed to freezing temperatures in soil of various moisture contents and with various periods of pre-chilling.

Variation due to:	Degrees of freedom	Sum of squares	Mean square	F
Varieties	5	5,025	1,005.0	4.31*
Moisture contents	2	5,979	2,989.5	12.82*
Freezing treatments	2	25,354	12,677.0	54.38*
Varieties x moisture contents	10	2,809	280.9	1.21
Varieties x freezing treatments	10	5,678	567.8	2.44**
Treatments x moisture contents	4	2,776	694.0	2.98**
Varieties x moisture contents x treatments	20	5,395	269.8	1.16
Error	162	37,770	233.1	-
Total	215	90,786	-	-

^{*} Exceed the 1% point.
** Exceed the 5% point.

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influenced by the amount of pre-chilling to which the plants were subjected. For the most part it was found that plants growing in unchilled soil were more sensitive to variations in moisture content than were those growing in chilled soil. The data presented in Table XI show that the least variation in survival occurred when the plants were pre-chilled for twelve hours. When the plants were pre-chilled for four hours, those on the soil having an intermediate moisture content gave relatively high survivals while those on the soil having a low moisture content gave relatively low survivals. Also, it will be noted that those plants which were not chilled gave relatively high survivals on soil having a low moisture content and relatively low survivals on soil having an intermediate moisture content.

It has been shown (6) that the temperature of moist soil lags more upon exposure to low temperatures than does that of dry soil. This protective effect of water can be used to explain the fact that, on the average, the plants growing in wet soil suffered less than similar plants growing in dry soil. It would be expected that, the differences in survival of plants growing on wet and dry soil would be lessened when pre-chilling occurred as the wet soil would have radiated much of its potential heat previous to the advent of freezing temperatures. In general, the results obtained indicate that this occurred. More important, however, from the standpoint of technique is the fact that comparable results cannot be obtained unless the moisture content of the soil is uniform throughout the experiment at

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the time when the plants are exposed to freezing temperatures. It is also worthy of note that pre-chilling for twelve hours reduces variability from this source and is, therefore, an added precaution that may be expected to help in reducing such an error.

Experiment 2.

Me thods.

In this experiment the plants were grown in sand and in soil maintained at various moisture contents during the growth of the plants, but at a uniform moisture content during the exposure to freezing temperatures. Marquis wheat was the only variety used.

The plants were grown in glazed crocks of one gallon capacity. Two series of nine crocks each were used. The substratum used in Series I was Edmonton loam, while in Series II it was washed sand. A complete nutrient solution was added to the sand cultures. This solution consisted of 5 c.c. of calcium nitrate, 5 c.c. of potassium nitrate, 2 c.c. of magnesium sulphate, 1 c.c. of potassium acid phosphate and 1 c.c. of ferric tartrate per litre. The ferric tartrate was added every two days.

Three series, each consisting of three sand and three soil cultures, were maintained at constant moisture levels; one at 30, one at 45, and one at 60 per cent of the moisture holding capacity of the substrate concerned.

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At the time of freezing all cultures were brought to 60 per cent of their moisture holding capacities. They were pre-chilled for twelve hours and then exposed to a temperature of -12°C for four hours.

Results.

In Table XIII a summary of the survival indices of plants grown in sand and in soil at various moisture contents, and exposed to freezing temperatures at a uniform moisture content, is presented.

An analysis of variance was calculated from the data obtained. It was found that the mean square for moisture contents could not be regarded as being significant. It may be concluded therefore that growing the plants at these various moisture contents did not significantly alter their survival values when exposed to freezing temperatures at a uniform moisture content.

These results indicate that where ordinary care is exercised in watering, the variations in moisture content during the growth of the plants between containers in a given experiment, would not be sufficiently great to cause any marked variation in the survival values of the plants when exposed to freezing temperatures.

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TABLE XIII

Survival indices of Marquis wheat seedlings when exposed to a temperature of -12°C. for four hours after being grown in sand and soil cultures maintained at various moisture contents.

1	Mois	ture conte	ent		Sand culture	Soil culture	Average
30% of	the	moisture	holding	capacity	21.0	22.8	21.9
15% "	n	n.	Ħ	n	20.7	18.8	19.8
50% "	11	**	17	1t	19.1	22.1	20.6

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0.00	200	7.17)	*	e#	19	48	195

Hardening.

Literature review.

It has been shown by many investigators, among whom might be mentioned Hill and Salmon (4), Martin (9), Salmon (14), and Tumanov (18), that the resistance of winter wheats to freezing temperatures may be greatly increased if they are previously exposed, for a considerable period, to comparatively low temperatures. Under field conditions winter wheats are normally subjected to natural hardening in this manner before the onslaught of severe cold weather. Salmon (14) has shown that, in the unhardened condition, winter wheat varieties differed little; whereas, in the hardened condition, they differed greatly in their resistance to artificially produced freezing temperatures. Furthermore, these latter differences were more closely correlated with the differences exhibited in the field than were the former.

In considering the seedling frost reaction of spring wheats it is apparent that, under field conditions long periods of hardening previous to the advent of sub-zero temperatures, do not occur. Nevertheless, some hardening probably does occur as comparatively low temperatures for varying periods of time usually precede the advent of sub-zero temperatures.

Peltier and Kiesselbach (13) appear to be the only authors to have investigated the effect of hardening on the frost reactions of spring grains. They studied the effect of

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continuous and intermittent hardening and found that the continuously hardened plants were more resistant to frost injury than were the intermittently hardened plants. They conclude that either method may be used in determining varietal differences.

Methods.

Eight spring wheat varieties grown in flats, were pre-chilled for 36, 24, 12 and 0 hours and then exposed to a temperature of -10°C. for four hours. Each wariety was replicated six times within each hardening treatment.

Results.

A summary of the data obtained with regard to the survival of spring wheat varieties pre-chilled for various periods of time and then exposed to sub-zero temperatures is presented in Table XIV. An analysis of variance was calculated from the data obtained, and this is presented in Table XV.

The data show that pre-chilling markedly increased the frost resistance of the varieties concerned. Furthermore, pre-chilling for 24 or for 36 hours does not appear to be any more effective than pre-chilling for 12 hours.

The mean square due to the interaction of varieties and treatments (Table XV) cannot be regarded as significant.

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TABLE XIV

Survival indices of spring wheat varieties pre-chilled for various periods of time and then exposed to a temperature of -10°C. for four hours.

		Surviv	al ind	e I	
Variety	Pre-chilled 36 hours	Pre-chilled 24 hours	Pre-chilled 12 hours	Pre-chilled 0 hours	Mean
Ceres	67.8	68.3	75.8	19.8	58.0
D.C. I-28-65	64.7	73.3	67.2	26.3	57.9
Red Bobs 222	68.3	71.8	65.5	24.3	57.5
Reliance	63.8	63.7	73.2	23.0	55.9
Canus	66.2	70.2	60.2	17.5	53.5
Garnet	50.3	52.3	63.5	9.8	44.0
Marquis	61.7	53.8	42.7	9.3	41.9
Reward	54. 0	48.7	35.0	16.8	38.6
Mean	62.1	62.8	60.4	18.4	50.9

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. 6	3.91	9.85	1.84	10.45	Ů 5 1 €
1. E	5.81	4.00	16.80	1.2	

TABLE XV

Analysis of variance of the survival indices of spring wheat varieties pre-chilled for various periods.

Variation due to:	Degrees of freedom	Sum of squares	Mean square	F
Varieties	7	10,888	1,555	3.73*
Freatments	3	67,876	22,625	54.27*
Varieties x treatments	21	5,267	251	-
Error	160	66,751	417	-
Total	191	150,782		-

^{*} Exceed the 1% point.

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This indicates that the relative varietal reaction was the same in each of the hardening treatments and consequently it may be concluded that, in determining relative varietal reactions to freezing temperatures, the amount of pre-chilling to which the plants are subjected is of little consequence. However, it was noted that the coefficient of variability was considerably lower when the plants were pre-chilled than was the case when the plants were not pre-chilled. Again, increasing the duration of the pre-chilling period for more than twelve hours did not reduce the variability from that obtained when the pre-chilling period was twelve hours. It is known that in the freezing chamber used great care must be exercised if a uniform temperature is to be maintained throughout the chamber. It is possible that when flats containing warm soil (unchilled) are placed in such a chamber, the rapid radiation of heat from the soil sets up air currents that militate against a uniform temperature within the chamber, which results in variable survival. When, however, the plants are pre-chilled for twelve or more hours previous to freezing, the temperature of the soil approaches more closely that of the freezing chamber, with the result that radiation of heat is less rapid and there is less tendency to set up violent air currents.

With these considerations in mind, it has been the practice since this experiment was conducted to chill all plants for 12 hours previous to an exposure to freezing temperatures.

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Border effect within flats.

It was thought desirable to determine whether or not plants growing near the sides of the flats differed in frost reaction from similar plants growing hear the centre of the flat.

The border rows of each flat used in the experiment on the effect of varying moisture contents of the soil on the resulting frost injury of plants growing therein, were seeded to Marquis wheat. One row of Marquis also occurred at random in one of the inside rows. It will be recalled that three freezing treatments were used in this experiment. The first lot was subjected to a temperature of -12°C. for four hours without any pre-chilling. The second was pre-chilled for four hours and the temperature was then lowered to -12°C. and maintained for three hours. The third was pre-chilled for twelve hours and then subjected to a temperature of -12°C. for four hours.

As there were 12 flats occurring in each freezing treatment, there were survival indices from 24 border and 12 inside rows of Marquis available for comparison within each freezing treatment. A summary of the data obtained is presented in Table XVI. The statistical significance of the differences in survival indices noted were judged by the "T" method described by Fisher (2) for evaluating the significance of differences in means of small samples. It will be noted that in no instances were the differences in survival indices obtained statistically significant.

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TABLE XVI

Survival indices of border and inside rows of Marquis wheat seedlings exposed to freezing temperatures.

		surviva		70
Freezing treatment	Border	Inside	Difference	value
Not pre-chilled. Subjected to a temperature of -12°C. for four hours.	22.5	26.8	4.3	>.4
Pre-chilled for four hours, temperature lowered to -12°C. and maintained for three hours.	43.1	41.3	-1.8	>.4
Pre-chilled for twelve hours and then subjected to a temperature of - 12°C. for four hours.	30.7	28.5	-2.2	>.4

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Succ	A.3-	0.00	7,05	Then set of 1214-50 to 124-50 to 124

It would appear in the light of these results that border effect in flats may be neglected in evaluating the frost reaction of wheat seedlings.

Endosperm development of the seed.

Peltier and Kiesselbach (13) have pointed out that wheat seedlings are most susceptible to injury in the three-leaf stage, and they postulate that these plants are less able to withstand frost injury because, at this time, the food reserves of the endosperm have reached the point of exhaustion. If this theory is correct it would seem likely that if the normal development of the endosperm were interfered with to any great extent, seedlings grown from such seeds would be more susceptible to frost injury than seedlings grown from normal seeds. Furthermore, if such a relationship exists it is important in artificial tests to use seed that has attained a uniform development of food reserves.

Me thods.

The increase plots of Marquis wheat grown at various places on the University farm in 1934 were all injured by frost except one small lot grown in the greenhouse. It was thus possible to obtain a supply of seed that, as far as could be determined, was uniform except that it varied in frost damage. Four samples of seed were selected. One lot was normal (unfrozen); a second lot

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was lightly frozen; a third severely frozen and a fourth normal with a portion of the endosperm artificially removed. The weights of 1,000 kernel lots of these different types of seed are presented in Table XVII.

The seeds as described were sown in flats. One lot of flats had previously been filled with Edmonton black soil and another lot with Fallis gray soil.*

Each of the four seed treatments was replicated

16 times: 8 times on the black and 8 times on the gray soil.

When the plants reached the two-leaf stage they were pre-chilled for 12 hours and then exposed to a temperature of -100c. for four hours.

Results.

A summary of the data obtained is presented in Table XVII.

The standard error of the difference between the average survival indices of any two treatments was calculated and found to be equal to 5.1 units; consequently, differences between treatments must exceed 10.2 units in order to be considered significant. The data show that differences of this magnitude were not obtained. These results would indicate that the amount of endosperm present in the seed has little influence on the frost reaction of the plants grown from each seed.

^{*} A description of these soil types is presented in a later section of this paper entitled "The reaction of differently nourished plants to freezing temperatures."

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TABLE XVII

The effect of differences in soil type and endosperm condition on the survival indices of Marquis wheat seedlings exposed to a temperature of -10°C. for four hours.

	weight per 1,000	Mear	survival	index
condition of the seed used	kernels of the seed used	Black	Gray soil	Average
Normal	35.5	77	66	72
Endosperm partially removed	24.7	73	67	70
Slightly frozen	31.2	79	66	73
Severely frozen	22.7	68	66	67

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The Reaction of Differently Nourished Plants to Freezing Temperatures.

Literature review.

It has been shown by several investigators (7,11,15) that, within limits, the osmotic pressure of the cell sap is intimately associated with the cold resistance of the plants concerned. It is also known (7) that the osmotic pressure of the cell sap can be modified by varying the nutrition of the plant. Several investigators have, therefore, studied the response of plants to low temperatures under varying nutritional conditions.

Patanelli (12) studied the effect of sodium, potassium and magnesium salts on plants exposed to freezing and to chilling temperatures. He concluded that resistance to cold was not related to the concentration of cell sap nor with its salt content, but rather with the amount of sugar retained during cooling.

Sellschop and Salmon (16) applied solutions of various salts to soil in which were growing peanuts, cowpeas, maize and cotton and observed the effects of chilling these plants. They found that potassium nitrate and potassium chloride exerted a protective effect, while calcium chloride, calcium nitrate, sodium chloride and sodium nitrate exerted a deleterious effect upon the survival of the chilled plants.

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Holbert (5), by using a portable refrigerating machine, was able to freeze fertilized and unfertilized corn plants growing in the field. Plants growing in well fertilized plots were found to be more resistent to frost in the seedling stage, and also at the time of ear formation, than were similar plants growing in unfertilized soil.

Magistad and Truog (7) were also able to accentuate the cold resistance of corn plants by the application of fertilizers. They found that such an application increased the osmotic pressure of the sap of young corn plants which in turn lowered its freezing point one to two degrees. They concluded that the greatest benefits to be obtained from such a fertilizer application are likely to occur on peat and sandy soils low in soluble salt content.

extensive series of experiments with tomato, potato, barley, rye, wheat and oat plants, using specially constructed glass-enclosed cool temperature chambers. Plants were grown with normal amounts of potassium, nitrogen and phosphorous, and with deficiencies and excesses of each of these elements. It was found that those plants suffering from a deficiency of potassium had a very low osmotic pressure and suffered most from low temperatures, while those having an excess of potassium or a deficiency of either nitrogen or of phosphorous had a higher osmotic pressure than normally nourished plants and suffered less by exposure to cold. An excess of nitrogen or of phosphorous did not greatly change the osmotic

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pressure from that occurring in normally nourished plants. They conclude that an adequate application of potassium salts will best protect against low temperatures.

This work of Schaffnit and Wilhelm (15) indicates that
the protective effect of fertilizers is due to the specific
action of a particular ion rather than the correction of an
unbalanced nutrient condition. As the work of earlier investigators
was done with soil cultures, it was not possible to determine,
except in a general way, whether or not the protective effects they
noted were due to a specific ion or to the correction of an
unbalanced nutrient condition. As a result much work remains to
be done before this question can be answered with certainty.

In the present investigation it was hoped to study this problem by adding various nutrients to two different soils varying rather widely in chemical composition. By this means the elements most likely to be concerned in increasing or decreasing cold resistance could possibly be determined, and these could then be studied in detail. Unfortunately the studies have not, as yet, advanced beyond the preliminary stage.

Methods.

Two soils, a gray soil collected at Fallis, Alberta, and a black soil collected at Edmonton, Alberta, were used in these experiments. A detailed comparison of these two types of soil has been presented by Wyatt and Newton (21). The gray soils are rather badly leached, of poor fertility, and are slightly acid in reaction. The black soils, on the other hand,

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have not suffered by leaching to any extent, are very fertile and are neutral to very slightly acid in reaction. According to the analyses of Wyatt and Newton, cited above, the gray soils compared with the black are low in nitrogen, phosphorous and calcium. Positive responses in plant growth were obtained by these investigators when any of the above elements were applied as a fertilizer to these gray soils, but the most marked results were obtained when a combination of nitrogen, phosphorous and organic matter was applied. Fertilizers added to the black soil used in this investigation usually failed to give any marked response in plant growth. When such a response is obtained it usually occurs following the application of phosphates. Wyatt and Newton do not present data on the potassium content of the black and gray soils. However, when they applied this element as a fertilizer little response was obtained, consequently it would seem probable that an adequate quantity of this element is present in both of these soils to produce normal plant growth.

In all, three experiments were conducted in which the frost reaction of plants growing in these two soil types was compared.

In the first experiment three varieties of spring wheat, three of barley and two of oats were grown in the black and in the gray soil. Each variety was replicated eight times in each soil type. One half (four replicates) of the plants growing in the black soil and one half of those growing in the gray soil, were frozen shortly after they emerged and the balance when they had

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reached the two-leaf stage. In the case of the former plants, the endosperm of the seed was only partially exhausted at the time of freezing and the plants were probably still receiving a good deal of their nourishment from this source.

In all cases the plants were pre-chilled for twelve hours and then exposed to a temperature of -10°C. for four hours.

In the second experiment only two varieties, Canus and Marquis spring wheat, were used.

The following chemicals were applied to each soil at the rate of 200 pounds per acre: potassium acid phosphate, sodium acid phosphate, potassium nitrate, sodium nitrate and calcium oxide.

The five treatments and the check on each soil gave a total of 12 flats for a single replicate, and as this was approximately the capacity of the freezing chamber, it was necessary to repeat the experiment to provide for replication. In all it was repeated six times over a period of several months and the reactions of approximately 1,000 seedlings in each treatment on each soil type were determined. In all cases the plants were chilled and frozen as in the first experiment.

In the third experiment only one variety, Marquis, was used. The plants were grown in flower pots. Four lots of material were grown. One lot was grown on normal black soil, one lot on fertilized black soil, one lot on normal gray soil and one lot on fertilized gray soil. Each treatment was replicated 20 times, and each replicate of each treatment

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consisted of approximately 15 seedlings. The fertilizer applied was a "complete" fertilizer containing potassium, nitrogen, phosphorous and sulphur. It was applied at the rate of 200 pounds per acre.

The plants were chilled and frozen as in the first experiment.

Results.

A summary of the data obtained in the first experiment with regard to the reaction of wheat, oat and barley varieties, when grown on black and gray soil and then exposed to freezing temperatures in the one- and two-leaf stages, is presented in Table XVIII. An analysis of variance was calculated from the data obtained, and this is presented in Table XIX.

It will be noted that, on the average, those plants growing on the gray soil were more severely damaged than those plants growing on the black soil. This difference is statistically significant as is shown by the significant mean square due to soil types, Table XIX. Furthermore, the mean square due to the interaction of stages of development and soil types may be regarded as significant. This indicates that the difference in survival depends upon the stage at which the plants were exposed to the freezing temperatures. The data presented in Table XVIII show that the plants grown on the gray soil, and exposed to freezing temperatures shortly after emergence, suffered less but not significantly less than similar plants grown on the black soil. However, when the

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TABLE XVIII

Survival indices of wheat, oat and barley varieties grown on black and gray soils and exposed to freezing temperatures in the one- and two-leaf stages.

		Me	ean sur	vival	inde	н	
		Black	soil		Gray	8011	
Variety	C.A.N.	C.A.N. 1-leaf stage 2-leaf stage Av.	2-leaf stage	Av.		1-leaf stage 2-leaf stage	AV.
Wheat: Canus Garnet Marquis	1316	82.3 67.0 71.5	88. 60.8 53.8	63.9 62.9 4	86.0 55.0 65.5	325 0.05 30.35	70.9 48.0 47.9
Barley: Treb1 0.4.C.21 Glabron	753 734 718	85 4 4 8 8 8 8 9 9 9 9 9 9 9 9 9 9 9 9 9 9	70.8 61.0 64.8	54. 55.3 4.	57.3 58.5 51.8	422 2.53 800	53.6 47.8 42.2
Oats: Victory Legacy	518	28.3	59.58 8.80	43.9	85.0 23.8	31.8 23.3	23. 4. 6.
Average		50.9	62.0	56.5	55.9	35.9	45.9

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TABLE XIX

Analysis of variance of the survival indices of wheat, oat and barley varieties grown on black and gray soils and exposed to freezing temperatures in the one- and two-leaf stages.

Variation due to:	Degrees of freedom	Sum of squares	Mean	F
Stages of development	1	634.55	634.55	1.92
Soil types	1	3,538.45	3,538.45	10.71*
Stages of development soil types.	1	7,672.55	7,672.55	23.22*
Varieties	7	21,428.06	3,061.15	9.26*
Varieties x stages of development.	7	7,162.44	1,023.21	3.10*
Varieties x soil types	7	570.34	81.48	-
Varieties x soil types x stages of development.	7	668.56	95.51	-
Error	96	41,971.80	330.49	-
Total	127	83,646.75	_	

^{*} Exceed the 1% point.

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plants in the two-leaf stage were exposed to frost those grown on the gray soil suffered much more than did similar plants grown on the black soil. The relative damage by freezing temperatures to these latter plants grown on the black and gray soil, is shown in Figure 2. As the plants exposed to freezing temperatures shortly after emergence were probably obtaining their nourishment largely from the parent seed and those exposed in the two-leaf stage largely from the soil solution, it would appear, from the results obtained, that the difference in survival noted between plants growing on the gray and those growing on the black soil is due largely to the relative composition of the soil solution rather than to the physical characteristics of the soils concerned.

The mean square due to the first order interaction of varieties and soil types, and that due to the second order interaction of varieties and soil types and stages of development cannot, in either case, be regarded as significant. This shows that the relative varietal reaction to freezing temperatures, in both the one- and two-leaf stages, was the same when the plants were grown on the gray soil as it was when the plants were grown on the black soil.

A summary of the data obtained in the second experiment with regard to the reaction of two wheat varieties to freezing temperatures, when grown on gray and black soil to which had been added various fertilizers, is presented in Table XX. An analysis of varience was calculated from the data obtained, and this is presented in Table XXI.

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TABLE XX

The effects of soil type and fertilizers on the survival indices of Marquis and Canus wheat seedlings exposed to a temperature of $-10^{\circ}{\rm C}$

		N	I e a n	3 m	FVIV	8 1	Mean survival index	×	
	BI	Black soil		ਲ	Gray soil	1	Y	Average	8 6
Treatment	Cenus	Canus Marquis Av.	AV.	Canus	Canus Marquis	AV.	Cenus	Canus Marquis	AV.
Check	68.7	68.7 56.8	62.8	51.9	51.9 45.7	48.8	60.3	60.3 51.3	55.8
Potassium acid phosphate	67.9	53.0	60.5	56.5	48.8	52.7	62.2	50.9	56.6
Sodium acid phosphate	62.1	46.1	54.1	53.3	48.4	50.9	57.7	47.3	52.5
Potassium nitrate	52.6	44.0	48.3	50.4	36.9	43.7	51.5	51.5 40.5	46.0
Sodium nitrate	57.5	43.1	50.3	58.4	47.6	53.0	58.0	58.0 45.4	51.7
Calcium oxide	55.0	44.7	49.9	49.1	38.8	44.0	52.1	52.1 41.8	47.0

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TABLE XXI

Analysis of variance of the survival indices of Marquis and Canus seedlings grown on variously fertilized black and gray soils.

Degrees of freedom	Sum of squares	Mean square	F
5	9,254	1,851	5.04*
1	4,306	4,306	11.73*
5	3,584	717	1.95
1	16,759	16,759	45.66*
5	163	33	-
1	506	506	1.38
5	955	191	-
5	32,005	6,401	-
547	200,957	367	-
575	268,489	-	-
	5 1 5 1 5 1 5 5 5	5 9,254 1 4,306 5 3,584 1 16,759 5 163 1 506 5 955 5 32,005 547 200,957	5 9,254 1,851 1 4,306 4,306 5 3,584 717 1 16,759 16,759 5 163 33 1 506 506 5 955 191 5 32,005 6,401 547 200,957 367

^{*} Exceeds the 1% point.

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The standard error of the difference between the mean survival indices of any two treatments was calculated and found to be equal to 2.76, consequently differences in excess of 5.5 may be considered significant. It will be noted that the average survival index of plants growing on soil that had received an application of potassium nitrate, or that of plants growing on soil receiving an application of calcium oxide, was significantly lower than that of plants growing on soil that had received an application of potassium acid phosphate or that of plants growing on unfertilized soil.

On the average of all treatments, the plants growing in the black soil had a significantly higher survival index than did those growing on the gray soil.

As the mean square due to the interaction of treatments and soil types cannot be regarded as significant, it may be concluded that the various chemicals added affected the survival of plants growing on the gray soil in the same manner as it did those growing on the black soil.

There was no indication of a differential varietal response as none of the mean squares due either to varieties and treatments, varieties and soil types, or varieties and treatments and soil types, can be regarded as significant.

A summary of the data obtained in the third experiment with regard to the reaction of Marquis wheat seedlings to low temperatures when grown on completely fertilized and normal black and gray soils, is presented in Table XXII. An analysis of variance was calculated from the data obtained and is presented in Table XXIII.

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TABLE XXII

The survival indices of Marquis wheat seedlings grown on fertilized and unfertilized black and gray soils after exposure to a temperature of -10°C.

		Mean	survival	index
Treatment	Black		Gray soil	Average
Unfertilized	62.4		48.6	55.5
Fertilized	71.5		40.3	55.9
Average	67.0		44.5	-

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TABLE XXIII

Analysis of variance of the survival indices of Marquis wheat seedlings grown on fertilized and unfertilized black and gray soils.

Variation due to:	Degrees of freedom	Sum of squares	Mean square	F
Soil types	1	10,125	10,125	9.23*
Treatments	1	3	3	-
Soil types x treatments	1	1,497	1,497	1.36
Error	76	83,341	1,097	-
Total	79	94,966	-	-

^{*} Exceeds the 1% point.

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From the data presented it is evident that the addition of the complete fertilizer to these soils had no appreciable effect on the frost injury sustained by plants growing therein. There is a suggestion that it increased this resistance when applied to the black soil and decreased it when applied to the gray soil, but this differential effect cannot be regarded as being statistically significant.

Discussion.

It was pointed out that when the plants were exposed to freezing temperatures shortly after emergence, those growing on the gray soil did not suffer any more damage than did plants growing on the black soil; but when the plants were exposed to freezing temperatures when they had reached the two-leaf stage those growing on the gray soil were much more susceptible to injury than those growing on the black soil. It was thought that this latter difference was probably due to the difference in the composition of the soil solutions concerned. By the addition of comparatively large quantities of various chemicals, these soil solutions have been variously modified. Despite these modifications the differences in the survival values of plants growing on these soils and exposed to freezing temperatures remains largely unchanged.

It would appear from the results obtained that this difference is not due to a deficiency in the gray soil of any of those elements commonly found to limit plant growth under field

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conditions. These results do not, however, preclude the possibility of the difference in survival being due to the chemical composition of the soil solutions. It is also possible that this difference in survival is due to the physical condition of the soils concerned. Modifications in the physical nature of these soils and the determination of the reaction to freezing temperatures of the plants growing therein has not been attempted. It may be possible that the physical nature of the soil affects the frost injury sustained by older plants, even though it apparently did not affect the frost injury of plants recently emerged.

The reason for the difference in survival of plants grown on the two soils and exposed to freezing temperatures, is not only of scientific interest but also of practical significance, as a knowledge of the factors involved might be of value in determining field practices that would tend to lessen the frost hazard. It would appear therefore that further work upon this problem would be well worth while.

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The Reaction of Wheat, Oat and Barley Varieties to Freezing Temperatures

Literature review.

Aamodt and Platt (1) tested the seedling frost reaction of Trebi barley, Red Bobs 222, Marquis, Reward and Garnet wheat, Victory and wild oats, by means of artificial refrigeration. Trebi barley and Red Bobs 222 wheat were found to be the most resistant to frost damage, while Victory oats and wild oats were the most susceptible.

Peltier and Kiesselbach (13) have also tested the frost reaction of several varieties of barley, wheat and oats.

They concluded that in order of cold resistence these crops might be ranked as wheat, barley and oats, and also that varieties within any of these crops differ materially in cold endurance.

Waldron (19) noted that under field conditions spring
wheat varieties differed materially in their resistance to
damage by freezing temperatures. Ceres was noted to be quite
resistant, while under similar conditions Hope was very susceptible.

Gregory and Beeson (3) noted, after a naturally occurring frost, that winter wheat varieties heading at this time differed in the amount of damage sustained by the young flowers. Purkoff and Michikoff were found to be resistant to such damage, while Red Chaff, Goens and Rudy were susceptible.

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Varietal reaction to freezing temperatures in the seedling stage.

Methods.

In these experiments determinations were made on the reaction of 32 wheat varieties, 15 oat varieties and 29 barley varieties to freezing temperatures in the seedling stage.

Two separate determinations were made on the reaction of the wheat varieties to freezing temperatures. They were grown in August 1935 and again in March 1936. In each instance the plants were pre-chilled for twelve hours, when they had reached the two-leaf stage, and then the former were exposed to a temperature of -8°C. and the latter to a temperature of -11°C. for a period of four hours. In each test the varieties were replicated six times, each replicate being made up of approximately 20 plants of each variety.

The reaction to freezing temperatures of the barley varieties was determined in the same manner as those of the wheat varieties except that the plants used in the first test were grown in December 1935 and those used in the second test in February 1936.

Only one determination was made on the reaction of the oat varieties to freezing temperatures. These varieties were replicated only four times. The plants were pre-chilled for twelve hours when they had reached the two-leaf stage, and then exposed to a temperature of -10°C. for four hours.

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Results.

A summary of the data obtained on the reaction of spring wheat varieties to two different freezing temperatures is presented in Table XXIV. An analysis of variance was calculated from the data obtained and this is presented in Table XXV.

The data show that distinct varietal differences in survival following exposures to freezing temperatures exist among the varieties of spring wheat tested. It will be noted that the majority of the more resistant varieties are the result of crosses between spring and winter wheats. Among the better known varieties found to be resistant occurred Red Fife, Canus, Reliance and Ceres. The early varieties tested, namely: Reward, Khogoh, Garnet and Ruby appear to be relatively susceptible. Huron and Hope were also found to be quite susceptible.

The mean square due to the interaction of varieties and temperatures is not significant (Table XXV). This shows that the relative varietal reaction was the same when the plants were exposed to a temperature of -8°C. as it was when the plants were exposed to a temperature of -11°C.

A summary of the data obtained on the reaction of the oat varieties exposed to freezing temperatures is presented in Table XXVI. An analysis of variance was calculated from the data obtained and is presented in Table XXVII.

A winter oat variety, Winter Turf, and a stock culture of Avena brevis were included in this test and each proved to be quite resistant. Victory and Leader were the most resistant of the

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TABLE XXIV

Survival indices of wheat varieties after exposure to temperatures of -8°C. and -11°C.

			Mean	survival	index
Variety	N.S.N*	C,A.N.	At -8°C.	At -11°C.	Average
Marquis x Kanred	I-28-139	-	93.3	68.5	80.9
Ridit x Ceres	I-33-36	-	93.2	6 8.6	80.9
Canus	I-28-114	-	89.2	72.2	80.7
Milturum 0.321	I-28-14	1415	80.0	74.8	77.4
Marquis x Kanred	I-28-137	-	93.2	60.3	76.8
Milturum 0.274	I-28-25	1628	84.7	68.3	76.5
Marquillo x (MarqKan.)	I-28-65	-	84.7	67.1	75.9
Red Fife	I-0-19	1515	83.5	64.7	74.1
Marquis x Kanred	I-28-39	-	78.8	68.5	73.7
Reliance	I-29-4	1498	84.8	62.4	73.6
Ceres	I-25-1	1263	83.2	64.0	73.6
Marquillo x (MarqKan.)	I-28-60	-	88.7	56.1	72.4
Preston	I-30-3	1635	82.2	61.5	71.9
Progress	I-29-7	1590	79.7	62.5	71.1
Hussar x Hard Fed- eration.	I-32-42	-	85.0	56.4	70.7
Marquis x Kanred	I-28-138	_	79.8	60.2	70.0
Marquillo x (MarqKan.)	I-28-64	-	82.7	56.9	69.8
Caesium 0.111	T-28-20	1256	82.3	56.0	69.2
Reliance x Hope	I-31-10	-	75.8	62.0	68.9
Red Bobs 222	I-0-18	1637	71.0	66.5	68.8
Early Triumph	T-25-12	1291	85.0	50.5	67.8
Renfrew	I-0-20	1514	75.5	58.9	67.2
Marquis	I-0-9	1621	73.0	58.2	65.6
Kitchener	T-0-5	1363	81.5	49.5	65.5
Reward	I-25-21	1509	68.5	56.9	62.7
Hope I Ceres	I-31-12	-	68.7	55.8	62.3
Huron	I-0-4	1344	75.7	48.0	61.9
H42 x Marquis	I-32-29	-	66.8	52.8	59.8
Hope	I-29-9	1615	73.3	44.7	59.0
Khogoh	I-34-14	-	66.7	49.3	58.0
Garnet	T-25-13	1316	69.7	46.2	58.0
Ruby	S-22-42	1511	59.5	36.0	47.8

^{*} University of Alberta Nursery Stock Number.

The standard error of the difference between any two averages is 5.64

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TABLE XXV

Analysis of variance of the survival indices of wheat varieties exposed to freezing temperatures.

Variation due to:	Degrees of freedom	Sum of squares	Mean square	F
Varieties .	31	21,817.0	703.8	3.68**
Temperatures	1	40,262.0	40,262.0	210.80**
Varieties x temperatures	31	5,574.5	179.8	-
Replicates*	10	17,677.5	1,767.8	9.26**
Error	310	59,214.5	191.0	-
Total	383	144,545.5	-	-

^{*} Made of replicates and the interaction of replicates and temperatures.
** Exceeds the 1% point.

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TABLE XXVI

Survival indices of oat varieties exposed to a temperature of -10°C.

Translation.	C + N	Mean
Variety	C.A.N.	survival index
Winter turf	291	53.0
A. brevis	-	48.8
Black Tartarian	188	36.5
Red Rustproof	514	32.7
Leader	199	31.3
Victory	518	31.0
Black Algerian	174	29.0
Banner	62	27.0
White Cross	601	22.8
Liberty	504	21.0
0.A.C. 144	39	16.8
Nider	643	15.5
Alaska	458	13.0
Gopher	14	10.5
Legacy	460	10.3

The standard error of the difference between any two means is 8.98.

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TABLE XXVII

analysis of variance of the survival indices of oat varieties exposed to a temperature of -10°C.

Variation due to:	Degrees of freedom	Sum of squares	Mean square	F
Varieties	14	9,392.9	670.9	4.16*
Replicates	3	11,182.4	3,727.5	23.11*
Error	42	6,776.1	161.3	-
Total	59	27,351.4	-	-

^{*} Exceeds the 1% point.

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varieties commonly grown in Alberta. The earlier varieties, particularly Nidar, Alaska, Gopher and Legacy were found to be quite susceptible to cold injury.

A summary of the data obtained on the reaction of the barley varieties to two different freezing tamperatures is presented in Table XXVIII. An analysis of variance was calculated from the data obtained and is presented in Table XXIX.

It was found that marked varietal differences in frost resistance exist among the barley varieties tested. Sacremento and Atlas appear to be quite resistant, while O.A.C. 21, Trebi, Regal and Glabron are among the varieties only slightly less so. Again, the early varieties, particularly Ottawa No.1014, Olli and Lapland were found to be quite susceptible. Two other early varieties, Pannier and Success, appear however to be moderately resistant.

The mean square due to the interaction of varieties and temperatures (Table XXIX) may be regarded as significant. This indicates that some of the varieties tested reacted in a differential manner when exposed to the two different freezing temperatures. Cross differences can be obtained between the responses of any two varieties to each of the two freezing temperatures. The differences between the mean survival indices of six replicates of each variety when exposed to two freezing temperatures were obtained by subtracting the survival index obtained at -11°C. from that obtained at -8°C. These data are presented in Table XXX. The standard error of the difference

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TABLE XXVIII

Mean survival indices of barley varieties after exposure to temperatures of -8°C. and -11°C.

		Mean	survival in	dex
Variety	C.A.N.	At - 8°C	At -11°C.	Average
Sacremento	744	74.0	55.2	64.6
Atlas	702	68.7	44.3	56.5
).A.C.21	1086	71.0	32.4	51.7
rebi	753	66.7	36.0	51.4
.A.C.21	734	64.0	34.6	49.3
legal	742	72.2	23.3	47.8
labron	718	68.0	27.0	47.5
colsess	772	72.0	22.5	47.3
lureka	773	57.0	35.6	46.3
uccess	783	64.2	25.8	45.0
eatland	722	62.0	24.4	43.2
Spartan	860	63.2	22.2	42.7
Taughn	1090	52.3	32.7	42.5
annier	1042	44.5	36.9	40.7
Tobarb	1022	47.8	28.2	38.0
old	829	59.2	15.5	37.4
Mannchen	837	60.3	13.7	37.0
Telvet	755	53.5	19.9	36.7
omfort	712	50.2	20.8	35.5
ttawa No.1014	1105	53.3	17.0	35.2
Himalayan	765	40.0	28.2	34.1
111	739	49.5	13.9	31.7
Fordon	833	45.3	17.3	31.3
Lapland	877	44.8	16.6	30.7
anadian Thorpe	816	49.0	10.3	29.7
Sanalta	1088	50.3	8.7	29.5
Bearer	704	30.2	19.4	24.8
Manchurian	724	27.3	20.8	24.1
Manchurian Newal	1089	27.5	11.1	19.3

The standard error of the difference between any two averages is 4.98.

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TABLE XXIX

Analysis of variance of the survival indices of barley varieties exposed to freezing temperatures

Variation due to:	Degrees freedom	Sum of squares	Mean square	F
Varieties	28	35,446.0	1,265.9	8.51**
Freezing temperatures	1	78,961.3	78,961.3	53.06**
Varieties x freezing temperatures	28	31,947.6	1,141.0	7.67**
Replicates*	10	108,945.2	10,894.5	73.21**
Error	280	41,677.6	148.8	-
Total	347	296,977.7	-	-

^{*} Includes replicates and the interaction of replicates and freezing temperatures.
** Exceeds the 1% point.

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TABLE XXX

Differences between the results of - 8°C. and -11°C. exposures according to survival indices.

Variety	C.A.N.	Difference
Sacremento	744	18.8
Atlas	702	24.4
0.A.C. 21	1086	38.6
Trebi	753	30.7
0.A.C. 21	734	29.4
Regal	742	48.9
Glabron	718	41.0
Colsess	772	49.5
Eureka	773	21.4
Success	783	38.4
Peatland	722	37.6
Spartan	860	41.0
Vaughn	1090	19.6
Pannier	1042	7.6
Nobarb	1022	19.6
Gold	829	43.7
Hannchen	837	46.6
Velvet	755	33.6
Comfort	712	29.4
Ottawa	1105	36.3
Himalayan	765	11.8
Olli	739	35.6
Gordon	833	28.0
Lapland	877	28.2
Canadian Thorpe	816	38.7
Sanalta	1088	41.6
Bearer	704	10.8
Manchurian	724	6.5
Newal	1089	16.4
Average		30.1

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between two such differences was calculated and found to be equal to 9.96 units, consequently differences in excess of 19.9 units may be considered significant. The average difference between all varieties exposed at -11°C. and at -8°C. was 30.1 units. It will be noted that Regal, Colsess, Pannier and Manchurian are among the varieties that gave a differential reaction. Such differential responses might well be expected. It would appear that the variety Regal, for example, is able to withstand a temperature of -8°C. quite satisfactorily but when it is exposed to a temperature of -11°C. the resistance that it exhibited at -8°C. was broken down, and the variety becomes relatively susceptible. On the other hand, the variety Manchurian is very susceptible even at -8°C. and consequently would not be expected to become increasingly susceptible at a lower temperature to the same extent as would other varieties which exhibited more resistance at the higher temperature. If a search for highly resistant varieties were being conducted, it would seem wise to use a freezing temperature that would severely injure the most resistant varieties in the test, and in this way it would be possible to eliminate those varieties whose resistance rapidly diminishes with a lowering of the freezing temperature. On the other hand, if it is desired to evaluate the actual difference between two varieties, it would seem necessary in conducting the test to use a range of freezing temperatures rather than one particular freezing temperature.

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Methods.

Two varieties of spring wheat, Marquis and Canus, were used in studying the reaction of wheat varieties to freezing temperatures at the time of heading. Four plantings of these two varieties were made with two days intervening between each planting. Within each planting the varieties were replicated ten times, each replicate containing about 30 plants of each variety. The plants were kept trimmed so that only one culm per plant was allowed to develop. At the time when freezing tests were to be made pollination had occurred in the first lot of plants sown, was just occurring in the second lot; the anthers were still green in the third lot, and the spike had not emerged from the boot in the fourth lot. As variation always occurs. even within a spike, these descriptions are indicative only of the stage of development of the majority of the spikelets concerned. As far as could be determined, the plants of one variety were comparable to those of the other in regard to the stage of maturity to which they had attained at this time. plants were pre-chilled for twelve hours and then exposed to a temperature of -4°C. for a period of four hours, except for two replicates of each planting which were retained as non-frozen checks. At maturity the number of fertile and non-fertile spikes was recorded.

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On August 21, 1935, a temperature of -3°C., of approximately three hours duration, occurred in the plots at this station. Immediately following this frost about 100 spikes of Canus and of Marquis were tagged. The anthers of these tagged spikes were still quite green, consequently they were somewhat comparable in their stage of development to the third planting referred to above. Unfortunately it was not possible to find spikes of these two varieties at other comparable stages of development. These tagged spikes were later harvested and the percentage of fertile and non-fertile florets per spike were noted.

Results.

A summary of the data obtained on the injury sustained by Marquis and Canus plants exposed to freezing temperatures at the time of heading are presented in Table XXXII. An analysis of variance was calculated from the data obtained and is presented in Table XXXII.

It was found that whenever a spike was damaged by artificially produced freezing temperatures, all florets on that spike were sterile, consequently the damage was noted as the percentage of spikes damaged by frost.

According to the data obtained, on the average of all determinations, Marquis suffered significantly more than did Canus.

The relative varietal reaction was the same, regardless of the stage at which the plants were exposed to freezing temperatures as is shown by the fact that the mean square due to the interaction of varieties and stages of development cannot be

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TABLE XXXI

The effect of exposure to a temperature of -40c. at different stages of development of Marquis and Canus wheat on the percentage of sterile spikes.

	Average	64.8	40.9		
Percentage of the spikes damaged by frost	Exposed shortly after the time of pollination	65.6	39.4	۵. « ه.	
	Exposed at the time of pollination	84.0	56.8	70.4	
	Exposed just previous to the time of pollination.	57.6	35.0	46.3	
	Exposed just previous to the emergence of the spike from the boot	51.9	32.5	42.2	
	Variety	Marquis	Canus	Average	

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TABLE XXXII

Analysis of variance of the percentage of sterile spikes of wheat varieties exposed to freezing temperatures at different stages of development.

Variation due to:	Degrees of freedom	Sum of squares	Mean square	F
Varieties	1	9,120.31	9,120.31	36.10
Stages of development	3	7,418.87	2,472.96	9.79
Varieties x stages of development	3.	155.32	51.77	-
Replicates	7	9,769.00	1,395.57	-
Error	49	12,379.00	252.63	-
Total	63	38,842.50	_	-

^{*} Exceeds the 1% point.

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considered significant.

It will be noted that the plants suffered significantly more damage when exposed to freezing temperatures at the time of pollination than they did when exposed at any other stage.

When the spikes of these varieties, exposed to naturally occurring freezing temperatures, were examined it was found that the type of injury was somewhat different than that obtained when the plants were damaged by artificially produced freezing temperatures. In these latter spikes it was found that in nearly all cases a part of the spike would be fertile while the remainder would be sterile. The sterile florets occurred apparently at random amongst the fertile florets. As a result the damage was noted as the percentage of sterile florets rather than as the percentage of sterile spikes. The upper and lower spikelets, which are commonly sterile, were discarded before the counts were made. It was found that the percentage of sterile florets was 58 and 51 for Marquis and Canus respectively. The statistical significance of the difference noted was examined by means of Fisher's T method. value obtained was greater than .2, consequently the difference noted cannot be regarded as being statistically significant.

The author is of the opinion that no conclusions may be drawn from these results until further data are obtained from other experiments of a similar nature. Circumstances beyond the author's control precluded the possibility of obtaining these data. The importance of frost damage at this stage of the plant's

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development, and the indications that varietal differences exist would seem to be sufficient grounds to warrant further work in this field.

Discussion.

The fact that the early varieties of wheat, oats and barley were found to be generally susceptible to frost injury suggests that a relationship may exist between these characters. In the experiments conducted all of the varieties in a given test were seeded at one time, and all the resulting plants were exposed to freezing temperatures at a given number of days after seeding. It is therefore possible that the early varieties were actually at a later stage of development when exposed to the freezing temperatures than were the later varieties. Then, if plants become more susceptible at later stages of development, the differences in survival noted would be at least in part due to this cause rather than to the inherent potentialities of the varieties concerned. It is, however, difficult to believe that the varieties differed in the stage of development that they had attained at such an early date after seeding, to a sufficient extent to account for the differences noted. Certainly any differences present were not noticeable to the eye.

If we assume that cold susceptibility is an inherent property of these early varieties, the suggestion at once presents itself as to the possibility of an association between factors for earliness and factors for cold susceptibility. In this connection

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it might be noted that Neatby (10) has shown that winter habit in barley is in reality the expression of an accumulated series of late growth factors. There would appear to be some grounds at least for assuming that winter habit is associated with cold resistance.

As earliness is, and cold susceptibility probably is, controlled by several genetical factors any association between these characters would probably be due to the multiple effect of single genes rather than due to a linkage between genes for earliness and those for cold susceptibility.

Any degree of association between these factors would tend to the production of early cold susceptible varieties and late cold resistant varieties, as no conscious selection has been made for cold resistance among the hybrid populations.

From the standpoint of erop improvement, this whole question deserves attention from the plant breeder. At present, considerable work is being done on the production of new early varieties of cereals. Even if it is granted that no association exists between earliness and cold susceptibility, if cold susceptible varieties are used as parental stocks, and no attention is paid to the cold resistance of the progeny, then there is a danger of obtaining varieties as susceptible or more so than those at present in use. As these new varieties are designed for those areas where frost injury is most likely to be a serious limiting factor in crop production, and as it appears that seedling frost damage seriously delays the maturity of the plants so injured, the danger involved

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appears to be sufficiently serious to warrant some attention being given to it. The knowledge of the approximate mode of the inheritance of cold resistance in cereal crops and more particularly of the associations, if any, between this character and any other characters would therefore be of considerable value to plant breeders who are developing crops for northern regions. Appende to an entrine play assume a present to the solution of the color of the color of the solution of the s

SUMMARY.

Five varieties of spring wheat were injured in the two- and in the five-leaf stage by frost and by defoliation.

One lot of this material was allowed to grow to maturity in the greenhouse and one lot was transplanted to the field, shortly after being exposed to freezing temperatures, and allowed to grow to maturity.

On the average, the lightly frozen plants were about three days later, the defoliated plants about five days later and the severely frozen plants about nine days later in heading than were comparable non-injured plants. The differences in days from emergence to heading were accentuated when the plants were grown to maturity in the greenhouse compared with the differences obtained when the plants were grown to maturity in the field. Those plants injured in the five-leaf stage were about two days later in heading than were plants injured to the same degree in the two-leaf stage.

The height of injured plants at maturity was found to be not greatly different from that of non-injured plants.

The number of fertile culms per injured plant was approximately the same as the number of fertile culms per non-injured plant when the plants were grown in the greenhouse, presumably under conditions of a very limited nutrient supply. When, however,

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the plants were grown in the field and having presumably a more generous nutrient supply, those plants severely injured by frost showed a reduction in fertile culms per plant of approximately 30 per cent, and those defoliated a reduction of approximately 20 per cent, when compared with non-injured plants.

Under normal field conditions plants defoliated in the two- and in the five-leaf stages were found to be significantly later in maturing, lower in yield per acreabut not different in height, when compared with non-injured plants.

There was no evidence that any one variety was consistently able to recover from the same degree of injury to a greater extent than were any of the other varieties tested.

Wheat plants were grown in soil maintained at approximately the same moisture content during the growth of the plants, but varied at the time the plants were exposed to freezing temperatures. The least injury occurred among plants growing in soil having a high moisture content and the greatest among plants growing in soil having a low moisture content. These differences were not as great when the plants were pre-chilled for twelve hours as they were when the plants were not pre-chilled.

wheat plants were grown in sand and soil cultures maintained at three different moisture contents during the growth of the plants but brought to the same moisture content at the time the plants were exposed to freezing temperatures. The survival indices of plants grown at the various moisture contents did not differ significantly.

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No significant differences were found to occur between the survival indices of plants growing in border and plants growing in inside rows of the flats used in these experiments.

The survival indices of plants grown from normal, slightly frozen, severely frozen, and seed from which a portion of the endosperm had been artificially removed, were found to be not significantly different.

Wheat, oat and barley plants were grown on a black highly fertile soil and on a gray relatively infertile soil. When these plants were exposed to freezing temperatures, shortly after emergence, those plants growing on the gray soil did not suffer any more injury than did those growing on the black soil; but, when the plants were exposed to freezing temperatures at the two-leaf stage, those growing on the gray soil suffered much more damage than did those growing on the black soil.

Potassium acid phosphate, sodium acid phosphate,
potassium nitrate, sodium nitrate and calcium oxide, were added
separately to these two soils. In no instance were the plants
grown in soils so fertilized more frost resistant than plants
growing in unfertilized soils. Furthermore, the addition of a
"complete" fertilizer containing the elements potassium, phosphorous,
nitrogen and sulphur, to these soils failed to increase the cold
resistance of wheat plants growing therein, as compared with those
growing in unfertilized soil.

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Wheat varieties were found to differ distinctly in their reactions to freezing temperatures in the seedling stage. Similar differences were noted among oat and among barley varieties.

Some evidence was obtained to show that wheat varieties may differ in their frost reaction at the heading stage.

Attention is called to the fact that the early maturing varieties of cereals tested in these experiments were all found to be relatively susceptible to frost injury in the seedling stage.

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